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Histological characteristics of sternoclavicular β2-microglobulin amyloidosis and clues for its histogenesis

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Histological characteristics of sternoclavicular β2-microglobulin amyloidosis and clues for its histogenesis.

Background. The pathogenesis of β2-microglobulin amyloidosis ($A\beta_2m$) has yet to be fully elucidated.

Methods. We describe the distribution and extent of $A\beta_2m$ deposition and macrophagic infiltration in cartilage, capsule, and synovium of sternoclavicular joints obtained postmortem from 54 patients after 3 to 244 (median 46) months of dialysis. Twenty-four nonuremic patients served as a control group. The diagnosis of amyloidosis (A) rested on a positive Congo Red staining (typical birefringence) and that of $A\beta_2m$ on positive immunostaining of the A deposits with a monoclonal anti-β2m antibody. The size of A deposits was measured.

Results. $A\beta_2m$ was detected in 32 (59%), and non-β_2m amyloid ($Anon\beta_2m$) was detected in an additional 8 (15%) of the 54 dialyzed patients. $A\beta_2m$ deposits were present in the cartilage of all $A\beta_2m$ (+) patients (100%). They were localized solely in the cartilage in 27% of the cases, either as a thin patchy layer or as a continuous thicker layer (identified as stage I). $A\beta_2m$ was additionally present in the capsule and/or synovium without macrophages in 27% of the cases (identified as stage II). The correlation between the size of cartilaginous deposits and dialysis duration ($P = 0.02$) as well as with the prevalence ($P = 0.03$) and size of capsular deposits ($P = 0.02$) suggests that stage II is a later stage of A deposition. Clusters of macrophages were detected around capsular and synovial amyloid deposits in 46% of the cases (identified as stage III). The longer duration of dialysis in those with stage III as well as the relationship between the size of the $A\beta_2m$ deposits and the prevalence of macrophagic infiltration suggests that stage III is the last stage of $A\beta_2m$ deposition. Marginal bone erosions were observed in 9 out of 12 patients with stage III deposits. Their size was correlated with that of cartilaginous deposits ($P = 0.01$). Among the 24 control patients, $Anon\beta_2m$ was detected in 12 patients (cartilage 100%, capsule 8%, synovium 30%).

Conclusions. The earliest stage of $A\beta_2m$ deposition occurs in the cartilage. $A\beta_2m$ subsequently extends to capsule and synovium. These two first stages do not require macrophage infiltration. Macrophages are eventually recruited around larger synovial or capsular deposits in the final stage. Marginal bone erosions develop in this late stage.

Key words: dialysis, histogenesis, sternoclavicular joint, macrophage, cartilage.

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The pathogenesis of β2-microglobulin amyloidosis ($A\beta_2m$), a major complication of dialysis, has yet to be fully understood [1, 2]. In a prospective study of joints obtained at autopsy, we recently reported histological evidence of joint $A\beta_2m$ in most patients within four years after the onset of hemodiaysis (HD) [3], prior to the clinical signs of the disease. We take advantage of the availability of whole joints to describe in more detail the distribution of $A\beta_2m$ and the extent of macrophage infiltration in the various components (cartilage, capsule, synovium) of sternoclavicular joints obtained in our autopsy study. We identify three stages of $A\beta_2m$ development: $A\beta_2m$ is first observed solely in the cartilage and subsequently extends to the capsule and synovium. Macrophages are not involved in these two stages. In the last stage, macrophages are recruited only around the larger synovial and capsular deposits, where typical marginal bone erosions develop.

METHODS

Dialyzed patients

All dialyzed patients in whom a large (diameter of more than 2 cm) sternoclavicular joint sample was obtained at postmortem were included in this study [3, 4]. Patients who had ever been given a renal transplant were excluded.

Fifty-four adult dialyzed patients (35 males) were studied, 34 of whom had been treated exclusively by HD, 16 exclusively by continuous ambulatory peritoneal dialysis (CAPD), and 4 by both methods. They had been on dialysis for 3 to 244 (median of 46) months. They were aged 42 to 91 (median of 64) years at the time of dialysis onset and 43 to 93 (median of 68) years at the time of death. Only two patients had undergone carpal tunnel syndrome surgery, one prior to the onset of dialysis.

The distribution of HD membrane types was similar to that reported in our recent study [3].

The etiology of end-stage renal disease was renal vascular disease ($N = 12$), chronic pyelonephritis or interstitial nephritis ($N = 12$), glomerulonephritis ($N = 8$), diabetic nephropathy ($N = 7$), polycystic kidney disease ($N = 6$), amyloidosis ($N = 3$), hemolytic uremic syndrome ($N = 1$), and unknown ($N = 5$).

The cause of death was myocardial infarction or heart failure ($N = 17$), peripheral or cerebrovascular disease ($N = 7$), infection ($N = 15$), cancer ($N = 7$), cachexia ($N = 3$), calciphylaxis ($N = 1$), chronic obstructive lung disease ($N = 1$), and unknown ($N = 3$).

Control patients

Twenty-four adult patients (12 males), aged 14 to 88 (median 73) years, without a history of uremia underwent postmortem sternoclavicular joint sampling. The cause of death was myocardial infarction or heart failure ($N = 12$), peripheral or cerebrovascular disease ($N = 3$), infection ($N = 5$), cancer ($N = 2$), graft versus host disease ($N = 1$), and autoimmune hepatitis ($N = 1$).

Joint specimens

All sternoclavicular specimens (one per patient) contained both a large cartilaginous and a large capsular sample and were thus considered representative. Most specimens [dialyzed patients, 44 (81%); controls, 21 (88%)] contained in addition synovium. All specimens were fixed in 4% buffered formaldehyde, decalcified in 5% formic acid for one to two weeks, embedded in paraffin, and cut into serial slides. The slides were stained by hematoxylin eosin and alkaline Congo Red. When detected, amyloid was typed by an avidin-biotin peroxidase complex and anti-β2m (dilution 1/100; Dako, Copenhagen, Denmark), anti-P component (dilution 1/500; Dako), anti-amyloid protein A (dilution 1/500; Dako), anti-prealbumin (dilution 1/200; Dako) anti-kappa (dilution 1/10,000; Bio-yeda, Rehovot, Israel), and anti-lambda (dilution 1/10,000; Bio-yeda, Rehovot, Israel) antibodies.

Macrophagic cells were stained by anti-CD68 (dilution 1/50; Dako) [5].

Diagnostic criteria and morphometry

The diagnosis of amyloidosis (A) rested on a positive Congo Red staining with characteristic green-yellow birefringence under polarized light.

When Congo Red staining was positive, immunoperoxidase stains were performed with anti-β2m and anti-P component antibodies. Whenever anti-β2m staining was negative ($Anon\beta_2m$), other antibodies were used [6].

The presence of macrophages was considered significant whenever a cluster of five anti-CD68 positive cells with a large cytoplasm was observed in the vicinity of amyloid deposits at a magnification of ×400 (Fig. 2).

β2m amyloid deposits were measured at a magnification of ×40 on anti-β2m immunostained slides with an eyepiece squared grid calibrated by a graduated picture on the glass surface of the slide. The grid has a surface of 1.69 mm^2 and is divided in 100. Each scale unit is 130 μm long and has a surface of 16,900 μm^2.

The size of β2m amyloid deposits in the cartilage was measured over five different fields, corresponding roughly to one third of the cartilage surface. The nonβ2m amyloid deposits were measured with the same technique on anti-P component immunostained slides. Capsular and synovial $A\beta_2m$ and $Anon\beta_2m$ deposits, unlike cartilaginous deposits, had highly variable sizes and shapes. Therefore, only the largest deposit was measured per high power field.

A bone erosion was defined as a loss of bone substance, replaced by connective repair tissue. Bone erosions were subdivided into two subgroups according to their location. Those located at the level of reflection of the synovial membrane on the cartilage (Fig. 3) were called marginal erosions, whereas those located at a distance from the reflection of the synovial membrane on the cartilage were called subchondral erosions. The surface of bone erosions was estimated as the product of their width by their length.

Hyperplasia of surface synoviocytes was considered significant when the number of cellular layers was five or more, even if hyperplasia was not uniform.

Statistical analysis

Results are presented as mean and SD, median and range, or percentage of patients at risk, meeting each criterion. Statistical analysis was performed by the chi-square test or the Fisher exact test for discrete variables and by the Mann–Whitney or Kruskal–Wallis test for continuous variables using the Epidemiology Program Office from the Center for Disease Control (Atlanta, GA, USA). Standard Pearson's linear correlation coefficients were also calculated. Differences were considered significant for a P of less than 0.05.

RESULTS

Dialyzed patients

Prevalence of $A\beta_2m$ and $Anon\beta_2m$. Among the 54 patients, $A\beta_2m$ deposits were detected in 32 (59%), and $Anon\beta_2m$ deposits were detected in 8 (15%; κ: $N = 1$, unknown $N = 7$). No amyloid was found in the 14 remaining patients (26%).

The three groups were comparable for age and gender distribution, but patients with $A\beta_2m$ had been on dialysis for longer than the two other groups (Table 1).

Tissue distribution of amyloid. Whenever amyloid, ei-

Table 1. Clinical characteristics of dialyzed and control patients

	Dialyzed patients					Control patients			
	$A\beta_2m$ (N = 32)	$Anon\beta_2m$ (N = 8)		No amyloid (N = 14)		$Anon\beta_2m$ (N = 12)		No amyloid (N = 12)	
Age at death *years*	69.5 (56–82)	67 (43–80)	NS	66 (53–93)	NS	74 (45–88)	NS	71.5 (14–88)	NS
Age at dialysis onset *years*	65.5 (46–76)	65.5 (42–78)	NS	61.5 (52–91)	NS	—		—	
Time on dialysis *months*	73 (17–244)	16 (3–44)	<0.001	43.5 (6–84)	0.015	—		—	

Results are presented either as median (range) or as prevalence (%) of patients at risk (N).
All statistics (P values) are by comparison with the $A\beta_2m$ group.

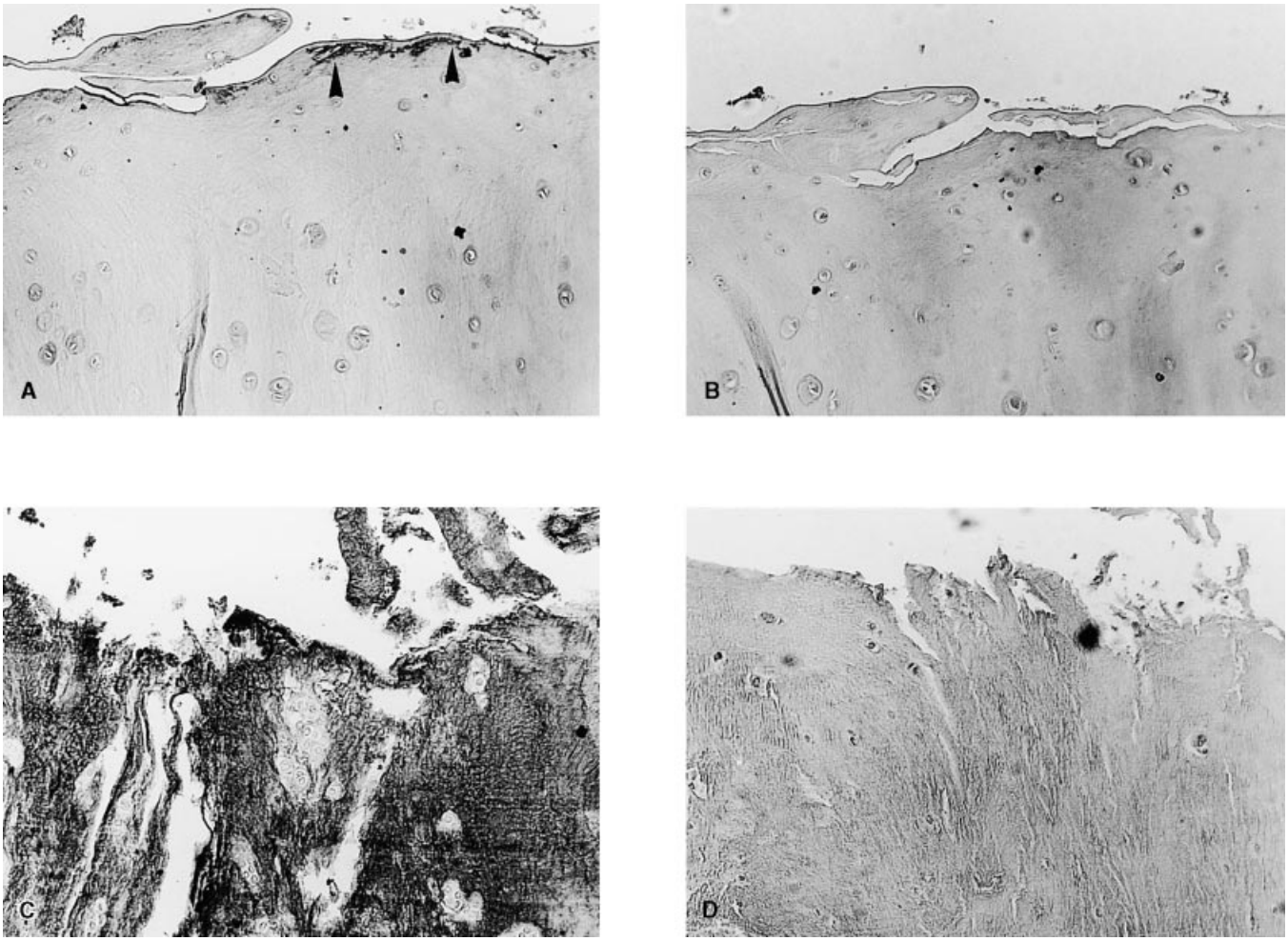

Fig. 1. Small cartilaginous $A\beta_2m$ deposits (arrowheads; *A* and *B*) and massive cartilaginous $A\beta_2m$ deposits (*C* and *D*); anti-β_2m immunoperoxidase (left) and anti-CD68 immunoperoxidase (right). There is an absence of macrophagic infiltration (magnification ×250).

ther of β2m or nonβ2m type, was detected in the joint, deposits were present in the cartilage. They were observed at least in the superficial layer of either intact or fissured cartilage (Fig. 1).

The size of $A\beta_2m$ deposits ranged from 0.03 to 0.95 mm^2 (median 0.152 mm^2). To assess the relationship between the size of cartilage deposits and several other pathological characteristics and duration of dialysis, joints with $A\beta_2m$ were divided in two groups: those smaller and those larger than the median value of 0.152 mm^2 (Table 2). Minor deposits (range of 0.03 to 0.11, median 0.05 mm^2) were often patchy, whereas larger deposits (range of 0.152 to 0.95, median 0.27 $mm^{2)}$ were more frequently (P = 0.004) continuous. The duration of dialysis was significantly longer in patients with large deposits.

Capsular (range of 0.03 to 0.8, median of 0.09 mm^2)

Table 2. Histological findings in dialyzed patients with small vs. large Aβ2m cartilaginous deposits

	Small	Large	*P* value
N	16	16	
Duration dialysis *months*	32.5 (17–160)	96 (24–244)	0.004
Deposits size mm^2	0.051 (0.034–0.118)	0.270 (0.152–0.946)	0.001
Continuous deposits in cartilage	31%	88%	0.004
Capsular deposits	38%	81%	0.03
Capsular macrophages	6%	25%	NS
Synovial deposits	38%	77%	NS
Synovial macrophages	23%	69%	0.047
Synovial cells hyperplasia	23%	38%	NS
Marginal bone erosions	23%	46%	NS
Subchondral cysts	38%	38%	NS

Results are presented either as median (range) or as prevalence (%) of patients at risk (*N*). Synovial samples were available only in 13 patients in both subgroups.

and synovial (range of 0.02 to 0.8, median of 0.2 mm^2) deposits were less frequent, both in the $A\beta_2m$ group (59 and 58%, respectively) and in the $Anon\beta_2m$ group (25 and 14%, respectively; Table 3 and Fig. 2). Capsular deposits were more frequent ($P = 0.03$) in the patients with larger cartilaginous amyloid deposits (Table 2).

The size of $Anon\beta_2m$ deposits in cartilage ranged from 0.017 to 0.169 (median 0.034) mm^2 and was thus much smaller than in $A\beta_2m$ ($P = 0.006$). The few $Anon\beta_2m$ deposits in capsule ($N = 2$) and synovium ($N = 1$) were small (≤ 0.1 mm^2).

Distribution and extent of macrophagic infiltration. Clusters of macrophages were never detected in the vicinity of $A\beta_2m$ or $Anon\beta_2m$ deposits in the cartilage. They were more often present around synovial (12 out of 15, 80%) than around capsular (5 out of 19, 26%) $A\beta_2m$ deposits ($P = 0.005$; Table 3). In the synovium macrophages were significantly more frequent around larger than around smaller $A\beta_2m$ deposits (9 out of 13 vs. 3 out of 13, $P = 0.047$; Table 2). Clusters of macrophages were never observed around $Anon\beta_2m$ deposits in capsule or synovium (Table 3). No significant infiltration of other inflammatory cells was observed around $A\beta_2m$ and $Anon\beta_2m$ deposits.

Bone erosions. Marginal bone erosions (range of 1.4 to 14.9, median of 5.8 mm^2) were observed in nine patients with $A\beta_2m$ and in none with $Anon\beta_2m$ (Table 4 and Fig. 3). They were characterized by a cystic dilation of the synovial reflection, including variable amounts of fibrous tissue with small vessels and macrophages. $A\beta_2m$ deposits were identified in eight of them. The nine patients with marginal bone erosions had been on dialysis for longer (34 to 244, median of 95 months) than patients with $A\beta_2m$ but without such marginal erosions (17 to 132, median of 31 months; $P = 0.02$).

Subchondral cysts were observed in 12 out of 32 (38%) patients in the $A\beta_2m$ group and 3 out of 19 (16%) in the $Anon\beta_2m$ group (Table 4). They were located under ulcerated cartilage as a bone erosion (with or without cavity). They included fibrous tissue, small vessels, some macrophages, fragments of altered bone, or cartilage. $A\beta_2m$ was detected in 7 out of 12 cases of the $A\beta_2m$ group. $Anon\beta_2m$ was found in one third of the cases of the $Anon\beta_2m$ group.

Synovial hyperplasia. Synovial hyperplasia was observed in 8 out of 26 (31%) patients with $A\beta_2m$, in 0 out of 7 (0%) patients with $Anon\beta_2m$, and in 1 out of 11 (9%) patients without amyloid (P = NS).

Relationship between the localization and size of amyloid deposits and time on dialysis. The size of cartilaginous $A\beta_2m$ deposits increases with the time spent on dialysis ($N = 32$, $r = 0.59$, $P = 0.01$) and is correlated with that of capsular deposits ($N = 19$, $r = 0.51$, $P = 0.02$).

The size of marginal bone erosions is correlated with the size of $A\beta_2m$ cartilaginous deposits ($r = 0.78$, $P = 0.01$ $N = 9$) but not with time on dialysis. The size of subchondral cysts is not correlated with the size of cartilaginous $A\beta_2m$ deposits or with the time of dialysis.

In $Anon\beta_2m$, no correlation was found between the size of cartilaginous deposits and the size of capsular deposits or time spent on dialysis.

Staging of joint amyloid deposition. Cartilage, capsule, and synovium were simultaneously available in 26 joints with $A\beta_2m$ (Fig. 4). $A\beta_2m$ was localized solely in the cartilage without macrophages in seven cases (27%). This stage was identified as stage I. $A\beta_2m$ was present additionally in the capsule and/or synovium without macrophages in seven more cases (27%). This stage was identified as stage II. Clusters of macrophages surrounded synovial and/or capsular deposits in 12 further cases (46%). This stage was identified as stage III. Marginal bone erosions were present in nine of the latter patients.

The size of cartilaginous $A\beta_2m$ increased from stage I (0.03 to 0.19, median of 0.03 mm^2) to stage II (0.05 to 0.9, median of 0.19 mm^2) and III (0.03 to 0.95, median of 0.27 mm^2; Kruskal–Wallis test for three groups, $P = 0.004$).

The time on dialysis increased from a mean of 39 [± 17 (SD) months] for stage I to 56 (±45 months) for stage II and 111 (±59 months) for stage III (Kruskal–Wallis test for three groups, $P = 0.003$). The difference between stage II and III does not reach statistical significance (Mann–Whitney test).

Control patients

The age distribution was similar in control and dialyzed patients ($P = 0.45$).

Twelve of 24 control patients had $Anon\beta_2m$ deposits [κ, $N = 1$, amyloid protein A (AA), $N = 2$, unknown type, $N = 9$]. They were older (median of 74 years) than the control patients without such deposits (median of 71.5 years), but the difference did not reach statistical

Table 3. Amyloid deposits and macrophages in joints of dialyzed and control patients

	Dialyzed patients			Control patients	
	$A\beta_2m$	$Anon\beta_2m$	No amyloid	$Anon\beta_2m$	No amyloid
Cartilage	$N = 32$	$N = 8$		$N = 12$	
Amyloid	100%	100%	—	100%	—
Macrophages around amyloid	0%	0%	—	0%	—
Capsule	$N = 32$	$N = 8$		$N = 12$	
Amyloid	59%	25% NS	—	8% 0.007	—
Macrophages around amyloid	26%	0% NS	—	0% NS	—
Synovium	$N = 26$	$N = 7$		$N = 10$	
Amyloid	58%	14% NS	—	30% NS	—
Macrophages around amyloid	80%	0% <0.001	—	0% <0.001	—

Results are presented either as median (range) or as prevalence (%) of patients at risk (N).
All statistics (P values) are by comparison with the $A\beta_2m$ group.

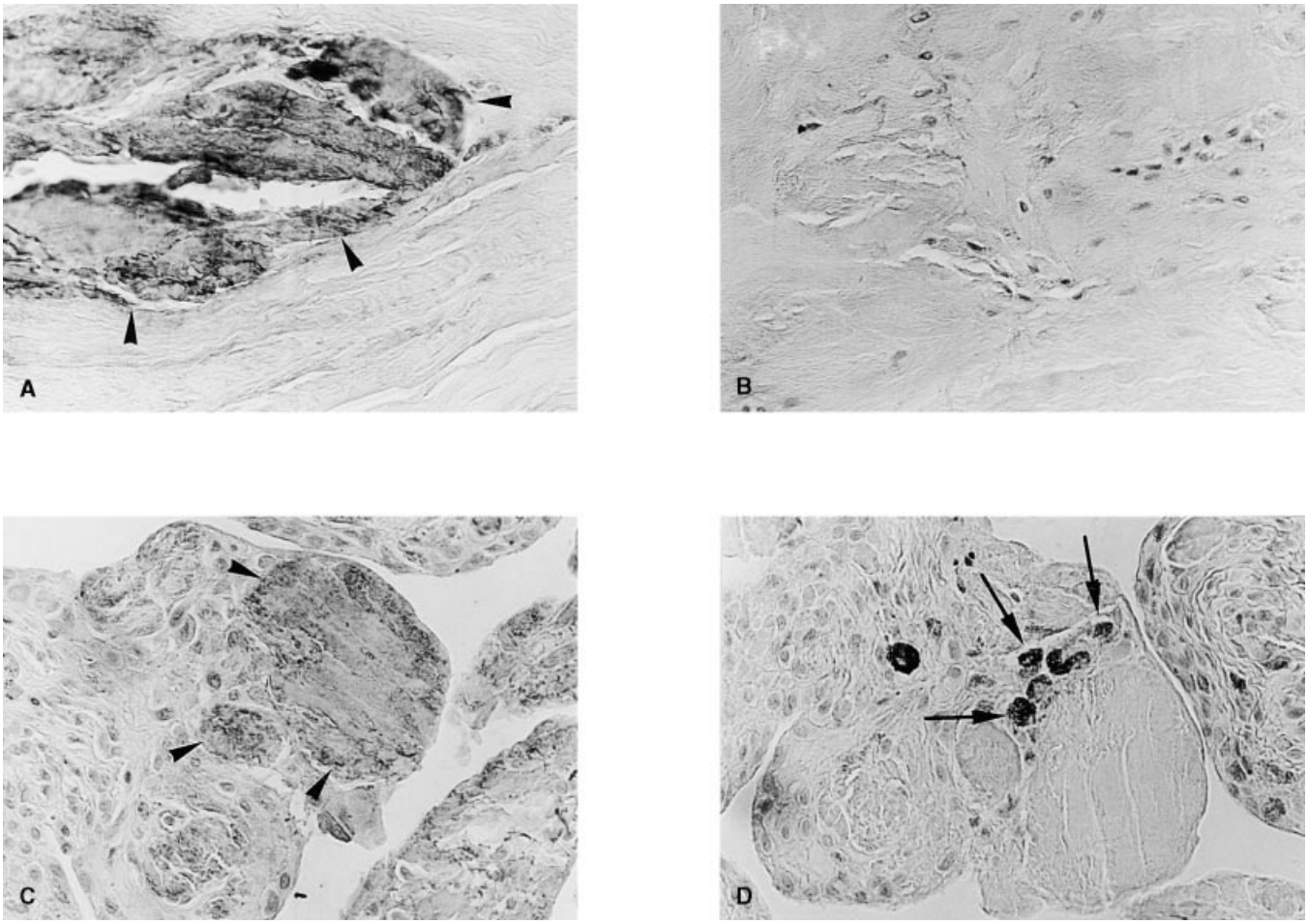

Fig. 2. Large capsular $A\beta_2m$ deposits (arrowheads) without macrophagic infiltration (*A* and *B*) and synovial $A\beta_2m$ deposits (arrowheads) with macrophages (arrows) in the vicinity (*C* and *D*); anti-β_2m immunoperoxidase (left) and anti-CD68 immunoperoxidase (right). (Magnification ×400.)

significance. $Anon\beta_2m$ deposits were present in the cartilage of all positive patients and were less frequently present in the capsule or the synovium (Table 3).

$Anon\beta_2m$ deposits in the cartilage were small (range of 0.017 to 0.152, median of 0.093 mm^2) and thus comparable to the deposits observed in dialyzed patients with small $A\beta_2m$ deposits (Table 3). Clusters of macrophages were never observed around $Anon\beta_2m$ deposits.

Marginal bone erosions were never observed, in contrast with subchondral erosions (Table 4). Synovial hy-

Table 4. Bone erosions in joints of dialyzed and control patients

	Dialyzed patients				Control patients		
	$A\beta_2m$	$Anon\beta_2m$		No amyloid	$Anon\beta_2m$		No amyloid
N	26	7		11	12		11
Marginal erosion	35%	0%	NS	0% 0.035	0%	0.039	0% 0.035
$N \rightarrow N$	32	7		12	11		12
Subchondral erosion	38%	43%	NS	0% NS	45%	NS	16% NS

Results are presented as prevalence (%) of patients at risk (*N*).
All statistics (*P* values) are by comparison with the $A\beta_2m$ group.

Fig. 3. Marginal bone erosion delineated by the arrowheads. Large $A\beta_2m$ deposits are present in the cartilage (arrows) and small $A\beta_2m$ deposits within the erosion (Congo Red, magnification ×20). C, capsule.

perplasia was observed in 0 out of 10 patients with $Anon\beta_2m$ and 3 out of 11 (27%) without amyloid (NS).

In control patients, stage III lesions were thus never identified, although stage I and II lesions were observed.

Comparison of dialyzed patients and controls

The pattern of amyloid deposition was similar in both groups. Whenever detected, amyloid was found in the cartilage and was less frequently found in capsule and synovium (Table 3).

Macrophages as well as marginal bone erosions were found in dialyzed patients only with $A\beta_2m$. By contrast, the prevalence of subchondral cysts and of synovial hyperplasia was similar in both groups (Table 4).

DISCUSSION

In this large study of sternoclavicular joint samples from dialyzed patients, we demonstrated that the presence of macrophages is not a prerequisite for $A\beta_2m$ deposition. Indeed, $A\beta_2m$ was deposited in the cartilage whether as a superficial, patchy layer or as a massive impregnation without detectable macrophages in their vicinity. Similarly, significant $A\beta_2m$ deposits develop in synovium or capsule in the absence of macrophages.

This finding is in contrast with current views implicating macrophages in the genesis of $A\beta_2m$ [5]. In amyloid protein, light chain derived (AL) and AA amyloidosis, the precursor protein is proteolyzed by macrophages prior to its deposition as an amyloid fibril [7, 8]. By analogy, it has been suggested that macrophages modified β2m as a first step in the precipitation of $A\beta_2m$ fibrils. Indirect evidence was provided by Morita et al, who detected β2m amyloid filaments within the cytoplasm of macrophage-like cells surrounding the amyloid deposits [9]. Subsequently, Knudsen et al demonstrated the synthesis and release of β2m by macrophages after cytokines or lipopolysaccharide stimulation [10], whereas Campistol et al reported the polymerization of β2m into fibrils in the supernatant of cultures of peripheral blood mononuclear cells harvested from dialysis patients [11]. More direct evidence for a central role of macrophages in β2m fibrillogenesis accrued from the observation that $A\beta_2m$ deposits were heavily infiltrated by macrophages [5]. In the latter studies, however, macrophagic infiltration was observed in only late (symptomatic) deposits of $A\beta_2m$ harvested during carpal tunnel syndrome (CTS) surgery ($N = 6$) or from an amyloid bone cyst ($N = 1$). In contrast, our study includes early preclinical $A\beta_2m$ deposits, many of which were confined to the cartilage. Thus, macrophages are not required for β2m amyloid fibrils formation. The presence of β2m within the cytoplasm of macrophages is thus best interpreted as a secondary phenomenon, representing a possibly beneficial, resorptive, catabolic process. Interestingly, Badman et al

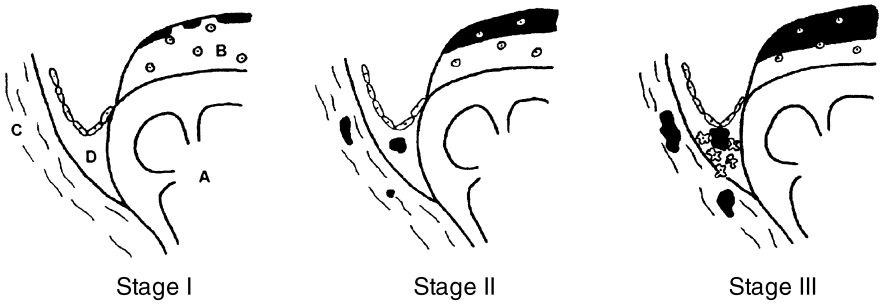

Fig. 4. Schematic representation of the three stages of $A\beta_2m$ deposition. $A\beta_2m$ is represented in black (bone *A*, cartilage *B*, capsule *C*, synovium *D*). Macrophages accumulate around synovial $A\beta_2m$ in stage III.

have recently demonstrated that pancreatic islet amyloid undergoes internalization by macrophages but resists proteolytic degradation [12].

More importantly, we demonstrated that macrophages are associated with a late stage of β2m amyloid deposition. Miyata et al have demonstrated that advanced glycation of β2m attracted macrophages, stimulated the release of proinflammatory cytokines, and contributed to bone resorption [13, 14]. This phenomenon might herald the transformation of clinically silent deposits into symptomatic amyloid masses with pain and bone cysts.

These data delineate three stages of $A\beta_2m$ deposition and might provide a scenario for $A\beta_2m$ progression in joints. The earliest sign of $A\beta_2m$ deposition is detected in the cartilage as a thin, superficial, patchy layer. Thereafter, the deposit increases in size and becomes continuous. The correlation between the size of the deposits and the duration of dialysis suggests a sequence between these two pictures, both of which are identified as stage I. Subsequently, $A\beta_2m$ appears in the capsule and/or in the synovium (stage II). These latter deposits are observed in association with only cartilaginous deposits. The prevalence and size of capsular deposits correlate with that of cartilaginous deposits. These two findings suggest that stage II represents a further step in joint $A\beta_2m$ accumulation. Macrophage recruitment around capsular and synovial deposits (stage III) appears to be a later phenomenon, as it is significantly more frequent in joints with larger $A\beta_2m$ deposits. It is at this stage that most previous histological observations have been collected [5].

Marginal bone erosions appear only at stage III. Their size is significantly correlated with that of cartilaginous $A\beta_2m$. The time on dialysis was significantly longer in patients with than without marginal erosions. These small erosions likely represent the early stage of the radiologically detectable marginal bone erosions characteristic of $A\beta_2m$ [1, 15]. In contrast, the prevalence of subchondral bone cysts is not different in dialyzed patients with $A\beta_2m$, $Anon\beta_2m$, no amyloid, and in controls. Clearly, subchondral cysts are not specific for $A\beta_2m$.

The various stages identified in this study reconcile various patterns of $A\beta_2m$ deposition. Athanasou et al previously reported the absence of macrophages around small $A\beta_2m$ deposits in the cartilage in an heterogeneous series of joint samples (vertebral, small and large peripheral joints) [16]. The size of the deposits was not quantitated, and macrophages were not detected by specific immunostaining. By contrast, Argilés et al reported large clusters of macrophages around massive $A\beta_2m$ deposits [5].

There are a few additional words of caution. The described sequence of stages remains hypothetical, as our study is only cross-sectional and does not provide sequential biopsies in the same patient(s). Furthermore, only sternoclavicular joints were included. Unlike most other joints, they contain mainly fibrocartilage rather than hyaline cartilage and are rarely symptomatic [17, 18]. They were nevertheless chosen because they are a frequent site of histological $A\beta_2m$ deposition [3, 4], and their small size allows the collection of all joint components in the same sample. Whether our findings do apply to other joints remains to be proven.

Nonβ2m (mainly senile?) amyloidosis was detected in 8 out of 54 dialyzed patients. This prevalence is certainly underestimated, as the identification of small nonβ2m deposits is almost impossible in the presence of larger $A\beta_2m$ deposits. It reaches 50% in the control group, in agreement with previous studies [19, 20]. This high prevalence underscores that diagnostic tests, such as the P-component scintigraphy [21], are not specific for $A\beta_2m$, as the P component should be taken up not only by $A\beta_2m$ but also by $Anon\beta_2m$ deposits. The latter include not only "senile" amyloidosis, but also, among others, AL [22] or AA amyloidosis that may coexist with $A\beta_2m$ (as observed in one patient in this series).

The amyloid deposition pattern is similar in $A\beta_2m$ and $Anon\beta_2m$. In both instances, it starts in the superficial layers of the cartilage and extends only later to other joint tissues. This similarity suggests that progress in the understanding of the pathogenesis of one type will contribute to the understanding of the other.

Several implications for future studies accrue from the current observations. First, the presence of $A\beta_2m$ in a sternoclavicular joint sample should not be excluded unless cartilage is available. Second, as early $A\beta_2m$ deposi-

tion does not require the participation of macrophages, further studies should concentrate on the cartilage as a nidus for $A\beta_2m$ fibril deposition. Cartilage alteration might account for two independent risk factors of $A\beta_2m$: time on dialysis and age. In this respect, Hou et al have recently proposed that the age-related collagen alteration is mediated by the accumulation of advanced glycation end-products [23]. Should this hypothesis be validated, new therapeutic interventions might be considered to inhibit advanced glycation with aminoguanidine or related compounds such as OPB 9195 [24, 25].

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